EUPHEM

EUPHEM REPORT

Summary of work activities

Baharak Afshar
European Public Health Microbiology Training Programme (EUPHEM), 2015 cohort

Background

According to Articles 5 and 9 of ECDC's founding regulation (EC No 851/2004) 'the Centre shall, encourage cooperation between expert and reference laboratories, foster the development of sufficient capacity within the community for the diagnosis, detection, identification and characterisation of infectious agents which may threaten public health' and 'as appropriate, support and coordinate training programmes in order to assist Member States and the Commission to have sufficient numbers of trained specialists, in particular in epidemiological surveillance and field investigations, and to have a capability to define health measures to control disease outbreaks'.

The ECDC Fellowship Training Programme therefore includes two distinct curricular pathways: Intervention Epidemiology Training (EPIET) and Public Health Microbiology Training (EUPHEM). After the two-year training EPIET and EUPHEM graduates are considered experts in applying epidemiological or microbiological methods to provide evidence to guide public health interventions for communicable disease prevention and control. Both paths that provide competency based training and practical experience using the 'learning by doing' approach in acknowledged training sites across European Union (EU) and European Economic Area (EEA) Member States.

European preparedness for responding to new infectious disease threats requires a sustainable infrastructure capable of detecting, diagnosing, and controlling infectious disease problems, including the design of control strategies for the prevention and treatment of infections. A broad range of expertise, particularly in the fields of epidemiology and public health microbiology, is necessary to fulfil these requirements. Public health microbiology is required to provide access to experts in all relevant communicable diseases at the regional, national and international level in order to mount rapid responses to emerging health threats, plan appropriate prevention strategies, assess existing prevention disciplines, develop microbiological guidelines, evaluate/produce new diagnostic tools, arbitrate on risks from microbes or their products and provide pertinent information to policy makers from a microbiological perspective.

According to the European Centre for Disease Prevention and Control (ECDC) Advisory Group on Public Health Microbiology ('national microbiology focal points'), public health microbiology is a cross-cutting area that spans the fields of human, animal, food, water, and environmental microbiology, with a focus on human population health and disease. Its primary function is to improve health in collaboration with other public health disciplines, in particular epidemiology. Public health microbiology laboratories play a central role in detection, monitoring, outbreak response and the provision of scientific evidence to prevent and control infectious diseases.

This report summarises the work activities undertaken by Baharak Afshar, cohort 2015 of the European Public Health Microbiology Training Programme (EUPHEM) at the National Infection Service, Public Health England, London, United Kingdom.
All EUPHEM activities aim to address different aspects of public health microbiology and underline the various roles of public health laboratory scientists within public health systems.

Pre-fellowship short biography

Dr Baharak (Bee) Afshar completed her first degree, in Biotechnology, at King's College London (KCL) University in in 1997. She was awarded a PhD on 'Diversity of *Mycoplasma fermentans'* at KCL University, in 2002. Baharak worked for a year as a Forensic Scientist at the Forensic Science Service in London before taking a post as a Project Scientist within Public Health England (PHE) and was involved in the development and evaluation of molecular methods for epidemiological typing of *Legionella pneumophila.* In 2008, Baharak was appointed as the DEVANI (Design of a Vaccine Against group B streptococcal Infections in Neonates) Project Scientist, which was funded through the European Commission. Baharak worked as a senior scientist on another research programme entitled 'Molecular anatomy of invasive group A streptococcal isolates in England' and worked closely with other project collaborators (Sanger Institute & Imperial College). She was appointed a permanent post taking a leading role in the development of novel molecular technologies for the key *Mycoplasma spp* and *Legionella spp.* national reference services within PHE, in 2014. During her EUPHEM fellowship Baharak has gained experience in virology, bacteriology, parasitology, epidemiology, bioinformatics and statistical analyis through a number of research projects based at Microbiology Reference Services, PHE Colindale, Field Epidemiology Service, PHE, Skipton House and the London School of Hygiene and Tropical Medicine.

Fellowship assignment: Public health Microbiology (EUPHEM) path

This report accompanies a portfolio that demonstrates the competencies acquired during the EUPHEM fellowship by working on various projects, activities and theoretical training modules.

Projects included epidemiological investigations (outbreaks and surveillance); applied public health research; applied public health microbiology and laboratory investigation; biorisk management; quality management; teaching and public health microbiology management; summarising and communicating scientific evidence and activities with a specific microbiological focus.

The outcomes include publications, presentations, posters, reports and teaching materials prepared by the fellow. The portfolio presents a summary of all work activities conducted by the fellow, unless prohibited due to confidentiality regulations.

Results

The objectives of these core competency domains were achieved partly through project or activity work and partly through participation in the training modules. Results are presented in accordance with the EUPHEM core competencies, as set out in the EUPHEM scientific guide[1].

1. Epidemiological investigations

1.1. Outbreak investigations

A. An international Salmonella Braenderup outbreak investigation using whole genome sequencing, March - June 2016

Supervisor: Lesley Larkin

Salmonella Braenderup is the 8th most common non-typhoidal Salmonella infection in England and Wales. *Salmonella* isolates submitted to the PHE Gastrointestinal Bacterial Reference Unit (GBRU) are routinely whole genome sequenced (WGS). A novel cluster of genetically closely related *S.* Braenderup cases reported from different geographical locations within Great Britain (GB) was identified by GBRU, in March 2016. By June, four other European countries reported an increase in this serovar.
A total of 56 isolates from England (45), Wales (2), Scotland (1), Jersey (1), Denmark (2), Sweden (2) Switzerland (2) and the Netherlands (1) were WGS and subjected to Single Nucleotide Polymorphism (SNP) analysis to confirm genetic relatedness. The 5-SNP single linkage cluster designation was 1.1.39.57.67.72.% with all isolates related at the 5-SNP level to at least one other isolate. Most isolates (85%) were indistinguishable at the core level, and over 50 SNPs from the nearest non-cluster *S.* Braenderup isolate. To generate a hypothesis for a potential food vehicle of

[1] European Centre for Disease Prevention and Control. European public health training programme. Stockholm: ECDC; 2017. Available from: http://ecdc.europa.eu/en/publications/Publications/microbiology-public-health-training-programme.pdf

infection, twenty trawling questionnaires were administered to randomly selected GB cases to identify common exposures which were investigated further using a targeted questionnaire on specific food items, shopping locations and loyalty card data. The outbreak was declared over by 26th June 2016 and the source of infection and vehicle(s) for transmission remain unknown. The fellow participated in contacting cases to complete the trawling and targetted questionnaires and attended subsequent outbreak control team meetings and prepared an outbreak report and presented this outbreak investigation at ESCAIDE, Stockholm, November 2016 and PHE annual conference, Warwick, March 2017.

B. Training modules

The EPIET/EUPHEM/FETP introductory course and the outbreak investigation module familiarised participants with the "10 steps of an outbreak investigation" and how to write an outbreak investigation report. This project was presented at the project review module in August 2016.

Educational outcome: Participation in national and international outbreak control meetings and teleconferences; questionnaire design and computer tools; involvement in outbreak investigation (case definition, interviewing cases and completing trawling and targeted questionnaires; collating and analysing data); collaboration and communication with PHE colleagues and health protection teams; application of combined microbiological and epidemiological knowledge in outbreak investigation; preparation of an outbreak investigation report and presentation at national and international conferences.

1.2. Surveillance

A. Surveillance of Mycobacterium tuberculosis (TB) cases attributable to relapse or reinfection in London, 2002-2015

Supervisor: Charlotte Anderson

The contribution of reinfection and relapse to TB incidence, and the factors associated with each are unknown. The main objectives of this study were to quantify and describe cases attributable to relapse or reinfection, and identify associated risk factors in order to reduce recurrence. The fellow performed a descriprive analysis as well as univariable and multivariable ananlysis using Stata. A first author manuscript is also in prepartion and will be submitted to *Epidemiology and Infection.*

B. Plasmid mediated quinolone resistance (PMQR) in carbapenemase producing Enterobacteriaceae (CPE) in the UK, 2014-2016

Supervisors: Matthew Ellington, Neil Woodford and Andre Charlett

There is a strong association between clinically significant fluoroquinolone resistance, contingent on chromosomal mutation, and the carriage of plasmids associated with multidrug resistant Enterobacteriaceae including those that encode extended-spectrum β-lactamases and carbapenemases (carbapenemase producing Enterobacteriaceae (CPE). The association between the carriage of plasmids encoding advanced β-lactamases and fluoroquinolones has been associated to plasmid-mediated quinolone resistance (PMQR) genes which confer a characteristic phenotypic minimum inhibitory concentration (MIC) profile, but the concordance between quinolone MICs and known PMQR genes (the phenotype-genotype concordance) amongst large isolate sets remains poorly probed raising the possibility that and as yet unidentified PMQR genes are circulating.
Between 2014-2016, 2,050 isolates from various species including: *Escherichia coli*, *Klebsiella* sp., *Enterobacter* sp., *Citrobacter freundii* were whole genome sequenced and analysed using in-house pipelines at the Antimicrobial Resistance and Healthcare Associated Infections (AMRHAI) Reference Unit based at PHE, Colindale. MIC data were also available for these isolates. The main objective of this project was to evaluate the ability of WGS to accurately predict plasmid mediated quinolone resistance in carbapenemase producing Enterobacteriaceae. Using Stata and Excel software, the fellow analysed the genotypic and phenotypic data from these 2,050 isolates in order to investigate the genotypic/phenotypic concordance in AMRHAIs fluoroquinolone susceptibility data for Enterobacteriaceae. This finding will contribute to the development of the WGS knowledge base and aid the translation of the technology into routine use. Patient management will be impacted by more accurately informed patient treatment, which in turn will minimise the likelihood of further resistance emerging. WGS based predictions of these mechanisms will enhance routine surveillance and improve our ability to track and interrupt spread of these mechanisms. The fellow is currently finalizing data analysis and preparing a manuscript for publication.

C. A retrospective surveillance study to determine the number of recurrent malaria cases and to genotype Plasmodium falciparum from these UK cases, 2008-2015

Supervisor: Colin Sutherland

Malaria is the tropical disease most commonly imported into the UK, with 1,300-1,800 cases reported each year, and 2-11 deaths. Approximately three quarters of reported malaria cases in the UK are caused by *Plasmodium falciparum*,

which is capable of invading a high proportion of red blood cells and rapidly leading to severe or life-threatening multi-organ disease. Artemisinin-based combination therapy (ACT) is recommended by the World Health Organization (WHO) as first-line treatment of uncomplicated *P. falciparum* malaria. The aim of this study was to identify potential examples of treatment failure in imported cases of Malaria to the UK from the dataset extracted from the PHE Malaria Reference Laboratory (MRL) database from 2008 to 2015, in order to evaluate and inform our national treatment guidelines. Using Stata, the fellow set up a search algorithm to identify two or more episodes occurring within a specific time period from the original treated episode (cut-off of: 6 weeks, 12 weeks, 24 weeks and 48 weeks), and identified eleven recurrent malaria cases within 2008 to 2015 dataset, this highlights the need to monitor and evaluate the efficacy of current drug regimen as a routine element of the management of imported malaria in the UK. The fellow is currently finalising data analysis and will prepare a joint manuscript with another EUPHEM (Laura Bubba, Cohort 2016) and FETP fellow (Nick Bundle) for publication.

Training modules

The EPIET/EUPHEM/FETP introductory course introduced the basic concepts of surveillance systems and building on this course the module on 'multivariable analysis' demonstrated the principles, application, and interpretation of multivariable analysis and provided a more comprehensive understanding of the principles of statistical analysis and how to build an optimal model using linear, logistic, Poisson and Cox regression in Stata.

Educational outcome: Understanding the challenges and limitations; formulation of specific public health recommendations; analysis of antimicrobial resistance genes derived from whole genome sequencing; univariable and multivariable analysis; preparation of descriptive report and writing of scientific articles.

2. Applied public health microbiology research

A. Mycobacterium tuberculosis (TB) cases attributable to relapse or reinfection in London, 2002-2015

Supervisor: Charlotte Anderson

The contribution of reinfection and relapse to TB incidence, and the factors associated with each are unknown. The aim of the study was to quantify and describe cases attributable to relapse or reinfection, and identify associated risk factors in order to reduce recurrence. Recurrent TB cases were categorised from notifications in the London TB register (2002-2015) as relapse or reinfection using molecular and epidemiological information. Demographic, social and clinical factors associated with each outcome were determined using logistic regression in Stata 13.1 (2009-2015 only). Of 43,465 TB cases analysed, 1.4% were classified as relapse and 3.8% as reinfection. Although the proportion of cases with relapse decreased from 2002 (2.3%) to 2015 (1.3%), the proportion of reinfection remained around 4%. Relapse was more common among recent migrants (<1 year), those with a social risk factor and those with central nervous system, spinal, miliary or disseminated TB. Reinfection was more common among long term migrants (>11 years), and those with a social risk factor within specific areas in London. The fellow performed a descriprive analysis as well as univariable and multivariable analysis using Stata. A descriptive report was produced by the fellow for internal (London PHE Health Protection Teams) and external stakeholders (TB Control Board, London TB Clinical Leadership Group and Local Authorities). Findings were presented to relevant staff working in TB control, decision makers including the London TB Control Board and London TB Clinical Leadership Group. The fellow developed a study protocol and has prepared a desriptive report and is currently preparing a manusript for publication to a peer reviewed journal. The fellow will also present the findings at national TB conference and international conference (ESCAIDE, Nov 2017).

Training modules

The EPIET/EUPHEM introductory course focused on the development and presentation of study protocols and the module 'initial management in public health microbiology' covered time management and team collaboration during projects.

Educational outcome: Preparation of research study protocol, descriptive report and an article; presentation to relevant staff working on TB control, decision makers including the London TB Control Board and London TB Clinical Leadership Group; findings will also be presented at ESCAIDE, November 2017.

3. Applied public health microbiology and laboratory investigations

A. Development of quantification and neutralisation assays for Zika virus

Supervisors: Catherine Thompson and Maria Zambon

The Zika virus outbreak in Brazil during 2015 has been linked to a rising incidence in microencephaly in babies born to mothers who were infected with Zika virus during pregnancy, this led to WHO declaring the South American Zika virus outbreak a Public Health Emergency of International Concern (PHEIC). Europe has responsibilities towards global Health to actively participate in projects to support research and development into the detection, diagnosis and spread of Zika virus infection and the link with microcephaly and Guillain–Barré syndrome (GBS). Currently, the standard assay for detection of Zika viral infection is a non-quantitative real-time PCR test that probes for the presence of viral RNA in clinical samples (i.e. serum, urine, amniotic fluid). However the effectiveness of this test is limited to a short period during the initial infection stage when Zika virus RNA is present, and cannot detect previous infection once the virus has been cleared. Hence, there is an urgent need to develop reliable, specific and cost-effective serological tests for Zika virus, in order to determine if patients, including neonates, have been exposed to the virus months earlier. The aim of this study was to develope and apply new methods for quantification of live Zika virus and viral RNA and a virus neutralisation assay to support serological testing from suspected or confirmed Zika virus infections.
This study comprised of three main objectives: (i) to develop, optimise and validate a new tissue culture infectious dose (TCID50) assay to quantify live Zika virus grown in tissue culture cells, (ii) to develop, optimise and validate a semi-quantitave RT-PCR assay to detect Zika virus RNA (iii) to develop and optimise a live virus neutralisation assay for Zika virus serological testing. The fellow has developed and optimised a new (TCID50) assay which is an ELISA-based method and a semi-quantitative multiplex real-time RT-PCR assay with an internal control. Standard opertaing procedures (SOPs) have been produced for these methods, however both assays require validation.

B. The first European multi-centre External Quality Assessment (EQA) study on phenotypic and genotypic methods used for Herpes Simplex Virus (HSV) drug resistance testing

Supervisor: Tamyo Mbisa

The main objectives of this study was to develop and coordinate the first European External Quality Assessment (EQA) study in order to evaluate phenotypic and genotypic methods used for HSV drug resistance testing in specialised reference laboratories from five centres in four European countries.
The fellow tested, prepared and dispatched an EQA panel of four HSV-1 or HSV-2 strains with different antiviral susceptibility profiles isolated from clinical samples. Isolates were quantified by qPCR, and aliquoted in culture medium. One isolate was distributed at two dilutions to help assess assay sensitivity. The panel was distributed to five European centres with a six-week deadline for the return of phenotypic and genotypic results, together with clinical reports. Four out of five participating laboratories returned results by the deadline. The fellow collated and analysed the results from all participating laboratories. Findings from this study were presented at a national meeting and published in *Journal of Clinical Virology.*

C. Preparation of Corynebacterium spp. panel and material for the WHO workshop

Supervisor: Androulla Efstratiou, Aruni de Zoysa and Gina Mann

The fellow examined, prepared and dispatched a panel of seven *Corynebacterium* spp. strains for the WHO workshop on 'Laboratory Dignosis of Diphtheria' for Cyprus, October 2017.

Training modules

The fellow received training on reference laboratory techniques for the detection and characterisation of potentially toxigenic *Corynebacterium* spp. Methods covered included culture and isolation on specialised media, confirmation and typing of species using biochemical tests, determination of toxigenicity status via the Elek test and detection of toxin genes by PCR. The fellow also received training on phenotypic and genotypic methods used for HSV drug resistance testing. Other methods included culture, TCID50 and neutralisation assays for Zika virus as well as formal introduction and assessment at the Virology Reference Department, PHE, for safe working in containment level 3 (CL3) laboratory.

Educational outcome: Preparation of SOPs for the following methods: (i) Detection of Zika virus using a multiplex real-time RT-PCR with an internal control assay and (ii) Determination of TCID50 of Zika Virus Stock Solution (ELISA based method); preparation of a PHE report on the international multi-centre EQA study and publication of a manuscript; presentations at national and international meetings; preparation of a report after the WHO workshop in Cyprus, October 2017.

4. Biorisk management

A. Preparation and dispatch of EQA panel of HSV-1 and HSV-2 strains

Supervisor: Tamyo Mbisa

The fellow prepared and dispatched HSV EQA panel to five participating centres from four European countries. HSV-1 and HSV-2 strains were sent on dry ice in compliance with current regulations (i.e. as biological substance, category B, packed to full UN3373 specifications).

B. WHO workshop: preparation a dispatch of Corynebacterium spp. cultures

Supervisor: Androulla Efstratiou

Corynebacterium diphtheriae, *C. ulcerans*, *C. xerosis*, and *C. striatum* cultures were tested, prepared and dispatched for the forthcoming WHO workshop on 'Laboratory diagnosis of Diphtheria' in Cyprus, October 2017. Cultures were sent on dry ice in compliance with current regulations (i.e. as biological substance, category B, packed to full UN3373 specifications).

C. Study on UK laboratory preparedness of emerging and re-emerging pathogens and diseases

In early 2016, the fellow did a research study on the 'UK laboratory preparedness of emerging and re-emerging pathogens and diseases'. The results were presented to the Director, the Chief Microbiologist, and the Chief Scientist of ECDC, during the Initial Management in Public Health Microbiology module at Stockholm, February 2016.

Training modules

The EUPHEM module 'Biorisk management' provided training on techniques used for both biorisk and biosafety assessment as well as mitigation, including WHO recommendations on biorisk management in laboratories. An overview of containment level 4 facilities was also provided and later complemented with a visit to two such labratories at the Public Health Agency of Sweden, Stokholm and at PHE in Colindale.

Educational outcome: Formal assessment and certificate was provided for international regulations on the transport of dangerous goods according to the International Civil Aviation Organisation (ICAO); practice of appropriate measures for the safe transport of hazardous substances and pathogenic specimens; understanding and experience of the principles and practice of biorisk management; knowledge of biosafety when working with infectious organisms; understanding of process associated with CL3 and CL4 laboratories; biorisk assessment and biorisk mitigation.

5. Quality management

A. The first European multi-centre External Quality Assessment (EQA) study for Herpes Simplex Virus (HSV)

Supervisor: Tamyo Mbisa

Herpes Simplex Virus (HSV) drug resistance is a significant public health concern among immunocompromised individuals. Phenotypic assays are considered the gold standard method for detecting HSV drug resistance. However, plaque reduction assays (PRAs) are technically demanding, often with long turnaround times of up to four weeks. In contrast, genotypic tests can be performed within a few days. The main objective of this study was to develop and coordinate the first European External Quality Assessment (EQA) study in order to evaluate phenotypic and genotypic methods used for HSV drug resistance testing in specialised reference laboratories from five centres in four European countries.
Four HSV-1 or HSV-2 strains with different antiviral susceptibility profiles were isolated from clinical samples. Isolates were quantified by qPCR, and aliquoted in culture medium. One isolate was distributed at two dilutions to help assess assay sensitivity. The panel was distributed to five European centres with a six-week deadline for the return of phenotypic and genotypic results, together with clinical reports. Four out of five participating labs returned results by the deadline. Phenotypic and genotypic data were largely, but not completely, concordant. An unusual resistance profile shown by one of the samples was explained by the detection of a mixed virus population after extensive further investigation by one of the centres. Discordant clinical outputs reflecting the diversity of phenotypic methodologies demonstrated the utility of this exercise. With emerging genotypic technologies looking to supplant phenotyping, there is a need for curated public databases, accessible interpretation tools and standardised control materials for quality management. By establishing a network of testing laboratories, this EQA scheme will facilitate ongoing progress in this area. The fellow was responsible for the overall management of this unique international multi-centre EQA study.

B. Laboratory audit

The fellow performed an internal audit of the National Legionella Reference Laboratory at PHE as part of the 'Quality Management' module.

Training modules

The EUPHEM 'Quality Management' module provided an overview of quality management concepts in diagnostic laboratories, according to the ISO 15189 standard. Topics covered included factors influencing quality in laboratories, internal and external quality control, accreditation, assessments and audits, documentation and record keeping, sample management, stock purchase and inventory management, management of equipment and temperature controlled devices, process improvement as well as customer service and international health regulations.

Educational outcome: Understanding the principles and practices of quality assurance; managing an international multi-centre external quality assurance study; designing questionnaires; contribution to an external accreditation audit; understanding local and European accreditation procedures.

6. Teaching and pedagogy

A. Bioinformatics and Phylogenetics Module for EUPHEM fellows

The fellow delivered a lecture at the very first 'Bioinformatics and Phylogenetics' module for the EUPHEM fellows from Cohorts 2014 & 2015. This module was hosted at the Folkhälsomyndigheten, Stockholm, November 2015. Title: "Whole genome sequencing of group A streptococci (GAS) *emm*/M3 isolates in England".

B. Organisation and management of the first workshop on gastrointestinal infections outbreak investigation

The Gastrointestinal Infections Department at PHE conducted the first workshop training session on outbreak investigation at Colindale over two days, Friday the 27th May and Wednesday the 1st June 2016. This workshop covered the steps in outbreak investigation from start to finish and focused on practical exercises interspersed with short presentations. The fellow organised and managed this workshop, developed the workshop programme, liaised with all presenters and participants, presented at the introduction and wrap up sessions. The fellow was also responsible for the workshop evaluation and preperation of certificates.

C. Organisation and management of the Lab4Epi Module

Lab4Epi is provided annually by PHE and has been described by Public Health England (PHE) as 'a training module intended to show the major role of public health laboratories in surveillance and outbreak investigations'. Its stated purpose has been to: 'improve the collaboration between epidemiologists and laboratory specialists building an understanding of perspectives and expectations'. The fellow co-organised this two-day module together with a fellow (James Elston) from the UK field epidemiologist training programme (FETP). In response to feedback from the previous Lab4Epi module and a learning needs assessment conducted with previous participants, the fellows modified the module objectives and programme for this year's course which took place in January 2017. The fellow also produced a Lab4Epi training module report.

D. Preparation and facilitation of a case study during the Lab4Epi Module

The fellow co-developed and facilitated a new case study on "Investigating a national outbreak of bacterial gastroenteritis in the era of whole genome sequencing" that was delivered during the lab4Epi module in January 2017.

E. WHO Workshop on Laboratory Diagnosis of Diphtheria

WHO global collaborating centre for Diphtheria, PHE and WHO regional office in Europe, Denmark are organising a three-day workshop on laboratory diagnosis on Diphtheria at the Medical School, University of Cyprus, Nicosia (11-13th October 2017). Participants from 12 countries of the Newly Independent States of the former Soviet Union will be attending this workshop. The main aim of the course is to strengthen the participants' skills and to build capacity in the laboratory diagnosis of diphtheria. The expected outcome of this workshop is to establish a formal coordinated approach to strengthen diphtheria surveillance.

The fellow has been involved in organising this workshop and will be taking part in demonstrating, teaching and facilitating this three-day workshop, including preparation and dispatch of strains, reagents and other consumables from PHE to Cyprus. The fellow will demonstrate various phenotypic and molecular techniques and will also present a lecture at this workshop, Title: "Molecular typing of *Corynebacterium diphtheriae* and *Corynebacterium ulcerans*: an overview", as well ad undertake a workshop evaluation.

7. Public health microbiology management

A. Public health microbiology management during project work

Public health microbiology management was an important component of all projects and activities during the fellowship. This included laboartory management; work flow management; consideration of ethical issues; team building and coordination and time management. Working with external collaborators and working in a

multidisciplinary team comprising microbiologists, clinicians, epidemiologists, bioinformaticians, statisitcians, government officials and public health officers.

B. Management during workshops and training module

The co-organisation of workshops and the Lab4Epi training module required good planning, organisation and communication skills. The fellow was involved in developing a new programme and inviting all the speakers and facilitators for the outbreak investigation workshop and Lab4Epi training module.

C. Management during an international outbreak investigation

During the international *Salmonella* Braenderup outbreak investigation the fellow participated in contacting UK cases in order to complete the trawling and targetted questionnaires and attended subsequent national and international outbreak control team meetings and prepared an outbreak report and presented this outbreak investigation at ESCAIDE, Stockholm, November 2016 and PHE annual conference, Warwick, March 2017.

Training modules

The one-week module entitled 'Initial management in public health microbiology' focused on the understanding of roles and responsibilities in public health management. Topics covered included the identification of different management styles, team roles and team evolution, prioritisation and delegation of tasks and the provision of structured feedback.

Educational outcome: Experience of working in a multidisciplinary public health team. i.e., liaising with health protection team members, clinicians, epidemiologists, microbiologists and bioinformaticians within PHE; understanding team management; understanding roles and formal responsibilities in public health microbiology; plan, schedule and organise research projects and meetings; communication through OCTs and presentations.

8. Communication

Publications

1. **Afshar B**, Bibby DF, Piorkowska R , Ohemeng-Kumi N, Snoeck R, Andrei G, Morfin F, Frobert E, Burrel S, Boutolleau D, Crowley B, Mbisa JL. 2017. A European multi-centre External Quality Assessment (EQA) study on phenotypic and genotypic methods used for Herpes Simplex Virus (HSV) drug resistance testing. *Journal of Clinical Virology* (manuscript accepted).
2. **Afshar B** and Efstratiou A. 2017. Chapter on "Streptococcal Diseases" for the new laboratory companion manual to the CDC Communicable Diseases Book (chapter accepted).
3. **Afshar B**, Carless J, Balasegaram S and Anderson C. Surveillance of *Mycobacterium tuberculosis* (TB) cases attributable to relapse or reinfection in London, 2002-2015 (in preparation).
4. **Afshar B**, Ellington M, Andre C and Woodford N. Plasmid mediated quinolone resistance (PMQR) in carbapenemase producing Enterobacteriaceae (CPE) in the UK 2014-2016 (in preparation).
5. **Afshar B**, Bubba L, Bundle N and Sutherland C. A retrospective surveillance study to determine the number of recurrent malaria cases and to genotype *Plasmodium falciparum* from these UK cases, 2008 to 2015 (joint-first authorship paper with another EUPHEM fellow, in preparation).
6. **Afshar B**, Turner CE, Lamagni T, Smith K, Al-Shahib A, Underwood A, Holden MTG, Efstratiou A, Sriskandan S. 2017. Enhanced Nasopharyngeal Infection and Shedding Associated with an Epidemic Lineage of *emm*3 group A Streptococcus. *Virulence*. May 1:0. doi: 10.1080/21505594.2017.1325070. [Epub ahead of print].
7. David S, **Afshar B**, Mentasti M, Ginevra C, Podglajen I, Harris SR, Chalker VJ, Jarraud S, Harrison TG, Parkhill J. 2017. Seeding and Establishment of *Legionella pneumophila* in Hospitals: Implications for Genomic Investigations of Nosocomial Legionnaires' Disease. *Clinical Infectious Diseases*. May 1;64(9):1251-1259. doi: 10.1093/cid/cix153.
8. Mentasti M, Cassier P, David S, Ginevra C, Gomez-Valero L, Underwood A, **Afshar B**, Etienne J, Parkhill J, Chalker V, Buchrieser C, Harrison TG, Jarraud S; ESCMID Study Group for Legionella Infections (ESGLI).2016. Rapid detection and evolutionary analysis of *Legionella pneumophila* serogroup 1 sequence type 47. *Clinical Microbiology and Infection*. Nov 30. pii: S1198-743X(16)30603-6. doi: 10.1016/j.cmi.2016.11.019. [Epub ahead of print].
9. David S, Mentasti M, Tewolde R, Aslett M, Harris SR, **Afshar B**, Underwood A, Fry NK, Parkhill J, Harrison TG. 2016. Evaluation of an Optimal Epidemiological Typing Scheme for *Legionella pneumophila* with Whole-Genome Sequence Data Using Validation Guidelines. *Journal of Clinical Microbiology*. Aug;54(8):2135-48.
10. Mentasti M, **Afshar B**, Collins S, Walker J, Harrison TG, Chalker V.2016. Rapid investigation of cases and clusters of Legionnaires' disease in England and Wales using direct molecular typing. *Journal of Medical Microbiology*. Jun;65(6):484-93.

Reports

1. Afshar B, Bibby DF, Piorkowska R , Ohemeng-Kumi N, Snoeck R, Andrei G, Morfin F, Frobert E, Burrel S, Boutolleau D, Crowley B, Mbisa JL. PHE report on: European multi-centre External Quality Assessment (EQA) study on phenotypic and genotypic methods used for Herpes Simplex Virus (HSV) drug resistance testing,

October 2016.
2. Afshar B, McCormick J, Kanagarajah S, on behalf of the outbreak control team. PHE report on: Investigation of an international outbreak of *Salmonella* Braenderup, March -July 2016, May 2017.
3. Afshar B and Anderson C. PHE report on: Surveillance of *Mycobacterium tuberculosis* (TB) cases attributable to relapse or reinfection in London, 2002-2015, June 2017.
4. Afshar B. Lab4Epi training module report, May 2017.

SOPs

1. Determination of TCID50 of Zika virus stock solution (ELISA-based method).
2. Detection of Zika virus using a multiplex real-time RT-PCR with an internal control (MS2) assay.

Conference presentations

1. "An international *Salmonella* Braenderup outbreak investigation using whole genome sequencing, March - June 2016", ESCAIDE Stockholm, November 2016 (Poster presentation).
2. "An international *Salmonella* Braenderup outbreak investigation using whole genome sequencing, March - June 2016", PHE annual conference, Warwick, March 2017 (Oral presentation).
3. "Zika virus: where are we now? Cause for concern?" Current topics in Paediatrics Infections conference, Cyprus, June 2017 (Oral presentation).
4. "Surveillance of *Mycobacterium tuberculosis* (TB) cases attributable to relapse or reinfection in London, 2002-2015", ESCAIDE, Stockholm, November 2017 (Oral presentation).

Other presentations

1. "Sequence-based typing (SBT) of *Legionella pneumophila*: results of the 10th ESGLI multi-centre SBT proficiency panel". ESGLI annual conference in London, September 2015 (Poster presentation).
2. "Current diagnostic methods for carbapenemase-producing enterobacteriaceae, Problem Based Learning". Introductory module, Spetses, September 2015 (Oral presentation).
3. "Whole genome sequencing of group A streptococci (GAS) *emm*/M3 isolates in England". Bioinformatics & Phylogenetics module, Stockholm, November 2015 (Oral presentation).
4. "UK laboratory preparedness of emerging and re-emerging pathogens & diseases". Initial Management in Public Health Microbiology module, Stockholm, Sweden, February 2016 (Oral presentation).
5. "A European multi-centre External Quality Assessment (EQA) study on phenotypic and genotypic methods used for Herpes Simplex Virus (HSV) drug resistance testing". HSV/HIV annual meeting, PHE, May 2016 (Oral presentation).
6. "GBS intrapartum screening tests". Expert Workshop on Group B Streptococcus, at the Department of Health, September 2016 (Oral presentation).
7. "EUPHEM – year one activities". ECDC site visit and mid-term evaluation of the fellowship, May 2016 (Oral presentation).
8. "Development of quantification and neutralisation assays for Zika virus". PHE meeting, January 2017 (Oral presentation).
9. "EUPHEM fellowship update". PHE, September 2017 (Oral presentation).
10. "Molecular typing of *Corynebacterium diphtheriae* and *Corynebacterium ulcerans*: an overview". WHO Workshop on Laboratory Diagnosis of Diphtheria, Medical School, University of Cyprus, Nicosia, October 2017 (Oral presentation).

9. International Missions

A. Rapid needs assessment at Elliniko refugee camps in Athens, Greece

Supervisor: Kostas Danis

In late June 2016, Médecins Sans Frontières (MSF-OCG) conducted a MMR vaccination campaign of the refugees (6 months to 15 year olds) in three camps in Elliniko. Before the campaign was launched, a rapid assessment survey was conducted on 24th June 2016 in those camps, to estimate the baseline vaccination coverage in children under the age of 15 years old, and assess the health and sanitary needs of the refugees in order to inform the provision of services better adapted to the needs and improve MSF support. The fellow visited all refugee camps in Elliniko which were separated into three sections including: Elliniko I (Hockey stadium) currently (source: Ministry of Migration as of 22/06/2016) houses approximately 1,286 refugees indoors as well as within rub halls, Elliniko II (Olympic arrivals) houses approximately 1,375 refugees indoors and elliniko III (baseball stadium) houses 951 refugees outdoors in tents. The refugees in these three camps mostly came from Afghanistan and spoke Farsi. The fellow was able to interview over 20 refugees and acted as a Farsi speaking translator. A total of 1,400 refugee children (<16 years of age) were vaccinated by July 2016.

Training modules

Training on how to carry out this task was given during the one-week module entitled 'Rapid Assessment and Survey' in Athens, June 2016.

Educational outcome: Undertaking rapid needs assessment at refugee camps; communication with refugees and project team members.

10. EPIET/EUPHEM modules attended

1. EPIET/EUPHEM introductory course, Spetses, Greece (three weeks)
2. ESCAIDE conference, Stockholm, Sweden, 2016 (three days)
3. Bioinformatics and Phylogenetics Module, Stockholm, Sweden (three days)
4. Outbreak Investigation Module, Berlin, Germany (one week)
5. Quality and Biorisk Management Module, Stockholm, Sweden (one week)
6. Initial PH Management Module, Stockholm, Sweden (one week)
7. Multivariable Analysis Module, Vienna, Austria (one week)
8. Rapid Assessment and Survey Methods Module, Athens, Greece (one week)
9. Project Review Module, Lisbon, Portugal, 2016 and 2017 (one week)
10. ESCAIDE conference, Stockholm, Sweden, 2017 (three days)

11. Other training and courses attended

1. Host site orientation and induction, Public Health England, London, UK (three weeks)
2. National conference on GBS, "Prevention of group B Strep infection in neonates: The way forward in the UK" for health professionals at the Royal College of Obstetricians & Gynaecologists, London, November 2015 (one day).
3. WHO certificate for International Transport of Infectious Substances, February 2016 (two days).
4. PHE *Legionella* day symposium: *Legionella pneumophila* (1976 to 2016) - from whole guinea pigs to whole genome sequencing, PHE, Colindale, March 2016 (one day).
5. Introduction to whole genome sequencing for foodborne public health microbiologists and epidemiologists, PHE, Colindale, April 2016 (one day).
6. Whole Genome Sequencing process and in-house pipelines used for sequence data analysis, Gastrointestinal Bacteria Reference Unit (GBRU), PHE, Colindale, May 2016 (one day).
7. Training in the containment level 3 (CL3) laboratory, Virology Reference Department, PHE, June 2016 (three days).
8. Annual symposium in Respiratory Infections organised by National Institute for Health Research (NIHR) – Imperial College London, South Kensington Campus, September 2016 (one day).
9. Training on how to medically validate routine Diphtheria test results, Respiratory & Vaccine Preventable Bacteria Reference Unit (RVPBRU), PHE, September 2016 (two days).
10. A National Infection Training in London organised by The Royal College of Pathologists, October 2016 (two days).
11. 4th Annual Reference Virology Symposium - Neurological Infections, PHE Colindale, January 2017 (one day).
12. Lessons Learned from Public Health and Clinical Incidents and Outbreaks, Harlow, January 2017 (one day).

Discussion

Coordinator's conclusions

One of the main goals of the EUPHEM programme is to expose the fellows to different public health experiences and activities, thus enabling them to work across various disciplines. This portfolio includes laboratory and epidemiological projects covering bacterial, parasitic and viral pathogens across a variety of disease programmes, such as vector-borne diseases, sexually-transmitted diseases, food and waterborne diseases, respiratory tract infections, vaccine-preventable disease and antimicrobial resistance. All projects here described were in line with the 'learning by doing' and 'on-the-job' training service approach of the EUPHEM programme and followed the core competency domains described professionals in mid-career and above.

Outbreak, surveillance and laboratory activities ranged from local outbreaks to the analysis of national databases or international mission to Greece. All contributed to the understanding of important public health issues. During the two-year fellowship, the fellow, supervisors and training site have demonstrated the capability of addressing communicable disease threats in a structured joint approach between public health microbiology and epidemiology such as the use of whole genome sequencing based SNP analysis as a tool for international outbreak investigation or epidemiology and surveillance of plasmid mediated quinolone resistance (PMQR) in carbapenemase producing Enterobacteriaceae. Projects involved different professional groups, such as physicians, laboratory technicians, epidemiologists, statisticians, government officials, public health officers and logisticians, strengthening the fellow's ability to work in a multidisciplinary team. The projects have been nicely selected to cover not only important public health topics such as Zika infection or drug resistances and malaria but also a very broad panel of microorganisms giving Baharak the opportunity to gain an outstanding training which we hope help her success in her future career

on Public Health Microbiology. Those activities were complimented by nine training modules providing theoretical knowledge. Projects had a clear educational outcome, with results communicated in scientific journals and at conferences.
The coordinator team concludes that the fellow has succeeded in performing all her tasks to a very high standard and with a professional attitude.

Supervisor's conclusions

The EUPHEM programme was a tremendous opportunity for Baharak and has provided her with the unique tools to find her niche in the field of public health microbiology and epidemiology whilst working with colleagues from very diverse disciplines, both nationally and internationally. This was a fantastic learning curve and opportunity also for the training site with Baharak being the first ever EUPHEM Member State track fellow to be appointed within PHE. For the host institution, it has also provided the opportunity to build new bridges and strengthen collaboration between the different sectors within public health on both a national and international level. It has been a pleasure to mentor Baharak for the past two years and it has been a sheer delight to see her develop within the programme and acquire many new skills, particularly within the fields of public health epidemiology, virology, parasitology and bioinformatics. Her projects covered all core domains within the programme and showed that she was able to work on these projects independently, only occasionally consulting colleagues and peers for advice. One of her greatest strengths is her desire to work internationally in the area of public health and she was given the opportunity to do this via the WHO workshop for Russian speaking countries on diphtheria. Baharak also presented her findings and outputs from her projects at various national and international meetings ranging from local PHE Meetings to International Congresses. Her scientific knowledge, excellent scientific and organisational skills and team spirit has been very much appreciated by all supervisors along with her open-mindedness, positivity, diligence and goal-oriented personality. It was a great pleasure to have Baharak as a EUPHEM fellow within PHE and we highly appreciate her contribution and achievements within the fellowship programme.
We wish Baharak every success for the future as she very much deserves a position within PHE or beyond that will make excellent use of the knowledge, tools and resources she has gained during the two year programme. I also hope that PHE/UK will find a solution to continue this very excellent programme despite Brexit! Public health microbiology is a discipline that is urgently needed within the UK and globally. Graduated EUPHEM fellows like Baharak would be excellent champions in the development of such a new programme on a global level.

Personal conclusions of fellow

The EUPHEM programme has provided a unique opportunity to receive professional training and experience in public health microbiology across a broad range of pathogens and disciplines. This prestigious programme has allowed me to be involved in diverse projects covering the different aspects of public health microbiology (bacteriology, virology and parasitology), from outbreak investigation, surveillance and applied public health microbiology research to the public health microbiology management, rapid needs assessment and teaching. This fellowship has empowered me to work in multidisciplinary teams from different levels within the public health system and has taught me how to prioritise, multitask and manage various activities as well as how to communicate, summarise and present studies concisely and effectively. The fellowship successfully bridges the gap between microbiology, epidemiology and bioinformatics by maintaining a close connection to the European Programme for Intervention Epidemiology Training (EPIET) and Field Epidemiology Training programme (FETP) networks. The structure of EUPHEM training with specific modules and "learning by doing" approach across different disciplines provided an excellent foundation to perform these tasks.
I have particularly enjoyed attending the training modules and getting to know the other fellows and experts from different fields and organisations across Europe and feel privileged to be part of this prestigious and growing EPIET/FETP/EUPHEM network which provides great basis for future collaboration nationally and internationally. I hope to continue to progress and apply my unique skills gained from this fellowship to help protect and improve people's health for many years to come within Public Health.

Acknowledgements of fellow

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